INVESTIGATION OF BIOPOLYMERS WITH THE HELP OF A NEW HIGHLY SENSITIVE PHOTOELECTRIC SPECTROPOLARIMETER

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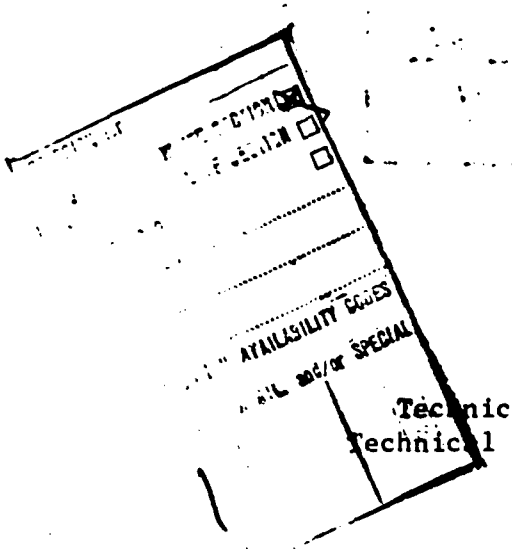

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INVESTIGATION OF BIOPOLYMERS WITH THE HELP OF A NEW HIGHLY SENSITIVE PHOTOELECTRIC SPECTROPOLARIMETER

/Following is the translation of an article by V. G. Alekseyev, M. A. Mokulskiy and V. I. Kurchatova, Institute of Atomic Energy imeni I. V. Kurchatova, published in the Russian-language periodical Biofizika (Biophysics) Vol X, No 2, 1965, pages 347-349. It was submitted on 3 Jan 1964. Translation performed by Sp/7 Charles T. Ostertag Jr./

At the present time investigations of the dispersion of the optical activity are finding wide application in various areas of science, especially in the study of the structure of high-molecular compounds.

In many cases it is necessary to investigate solutions of a low concentration, and consequently to conduct measurements of very small angles of optical rotation. Such a necessity may be connected with the non-linear dependency of the properties of the solution on the concentration, with a great deal of light absorption, and sometimes (in particular, when studying many biologically important compounds) with the difficulty of obtaining substances in large quantities. In such cases for the measurement of optical rotation with an accuracy of 0.001°, polarimeters with photoelectric recording are used /1--3/.

We have developed and constructed a new, more highly sensitive, photoelectric spectropolarimeter, intended for measuring the dispersion of optical activity in the area from 400 up to 700 mμ. As an extended operation test showed, this spectropolarimeter makes it possible to conduct measurements with microquantities of optically active substances, which is especially important for biological, chemical and physico-chemical investigations.

Table 1 presents data on the sensitivity of the device in various sectors of the spectral interval.

Table 2 presents data relative to the sensitivity of the device at various optical densities of the specimen being investigated.

The data presented in Table 2 was obtained when measuring in the green area of the spectrum with an aperature width of 40 Å.

For the purpose of determining the minimum concentrations of substances which are necessary for a reliable measurement of the optical rotation, we conducted an investigation of the dispersion of the optical activity of saccharose in water. These data are cited in Table 3. Sensitivity is expressed in seconds.

Measurements were conducted in a cuvette with a thickness of 50 mm.

We began investigations of complexes of nucleic acids with dyes of the acridine group. The interest in the investigation of complexes of acridine dyes with nucleic acids is caused by the fact that these substances possess biological activity (they may exert mutagenic effects, they are cancer producing, they are capable of inhibiting the synthesis of RNA on a DNA matrix and the replication of DNA). The first work on the investigation of these complexes by the method of dispersion of optical activity was performed by Bradley /4/. However, in investigating the complex of denatured DNA with acridine orange, Bradley did not detect any rotation connected with the dye. We conducted similar tests.

The diagram depicts the curves of dispersion of the optical activity of complexes of denatured DNA with acridine orange at a various concentration of nucleic acid and a constant concentration of dye.

As is seen from the diagram, the complex of DNA with acridine orange possesses an optical rotation in the adsorption band of the dye. With a change in the concentration of nucleic acid and a constant concentration of dye, the dispersion of optical activity is changed strongly, which apparently serves for a while as the cause of error, since in several proportions of the amount of polymer and dye the effect almost completely disappears.

The device assuredly measures optical rotation, with a maximum sensitivity of not exceeding 5". Measurements of the dispersion of optical activity of complexes are carried out in the adsorption band of the dye. The optical density of the samples in the interval from 460 to 500 mμ amounts to 1.5.

Literature

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b. Burer, T., Kohler, M., Gunthard, H. H., Helv. chim. Acta, 41, 2216, 1958.

c. Kuzel, V. A., Permogorov, V. I., Optics and Spectroscopy, 10, 541, 1961.

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Table 1

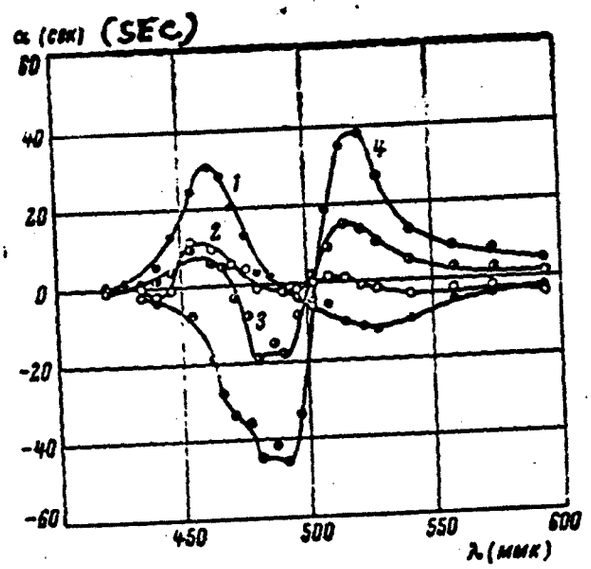
	Wavelength,						
	600	550	500	450	430	415	400
Sensitivity of device, sec.	1	0.8	0.8	1	1.5	2	3
Spectral width of aperture (Å) when measuring	60	45	30	27	24	17	12

Table 2

Optical density	0	0.5	1.0	1.5	2.0
Sensitivity of device, sec.	0.8	1	1--2	2--3	5--7

Table 3

Concentration of saccharose in solution, g/ml	Wavelength,				
	600	550	490	450	400
$2.25 \cdot 10^{-4}$	24	31	39	45	52.4
$1.123 \cdot 10^{-4}$	12.6	15.5	20	21.5	27.0
$0.562 \cdot 10^{-4}$	6.7	7.8	10.5	11.0	12.0



Dispersion of optical rotation of complexes of denatured calf-thymus DNA with acridine orange.

The natural rotation of DNA has been calculated. Concentration of dye $0.7 \cdot 10^{-5}$ M;

Concentation of DNA: 1 - $0.5 \cdot 10^{-5}$ M; 2 - $1 \cdot 10^{-5}$ M; 3 - $2 \cdot 10^{-5}$ M;

4 - $6 \cdot 10^{-5}$ M.